

# Characterization of Survival and Growth Effects Observed in *Hyalella azteca* Exposures to Fly Ash

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## INTRODUCTION

- Several independent studies (10–d and 28-d) indicate that *Hyalella azteca* exposures to fly ash result in reduced ( $\approx 25\%$ ) survival and growth
- Chemical constituents of fly ash (primarily metals/metalloids) are often assumed to be the cause of effects without confirmation
- Metals/metalloids are present in ash at concentrations that are marginal relative to sediment quality criteria for *H. azteca*
- Physical properties of fly ash (grain size and shape) are unlike those of natural sediment, so ash may not be a suitable habitat substrate for *H. azteca*
- No studies performed to date to discern causes of effects related to this release
- Previous unrelated studies have demonstrated the utility of separating porewater from whole sediment samples to characterize toxicity
- Guidance for sediment toxicity identification evaluation (TIE) studies describes laboratory techniques for characterizing and identifying toxicants in porewater and whole sediments

## OBJECTIVES

- Distinguish effects observed from *H. azteca* exposures to fly ash that are caused by physical properties of ash as opposed to metals/metalloids
- Characterize, individually, the bioavailability of metals/metalloids in whole ash and in porewater

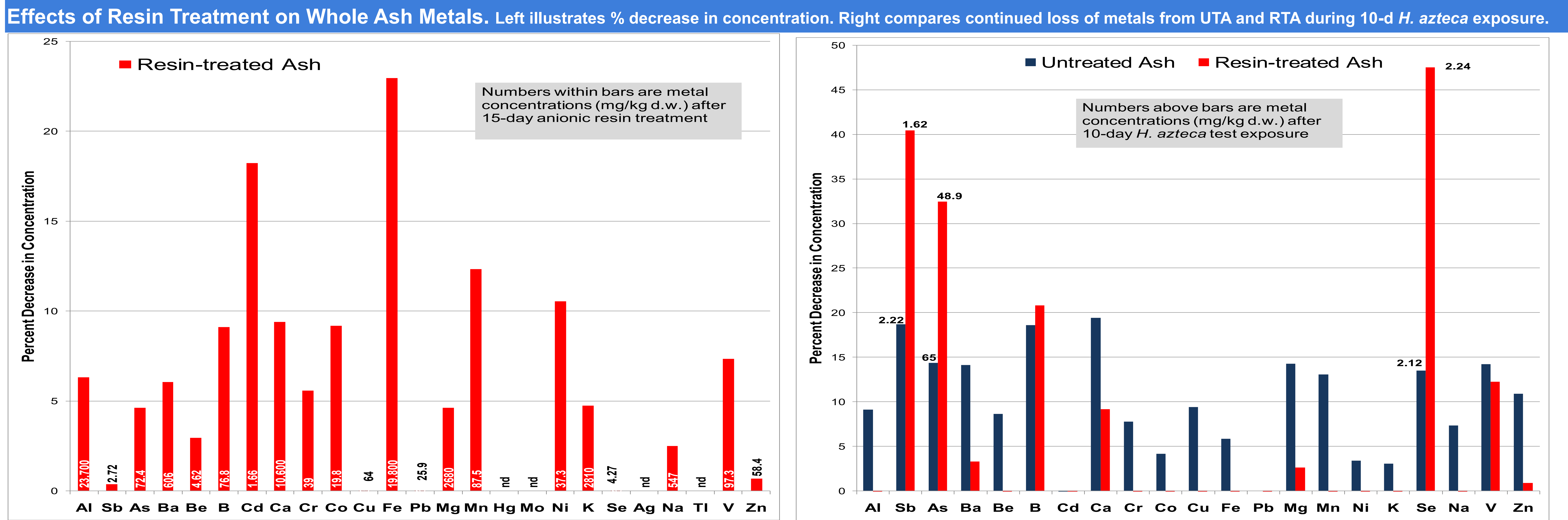
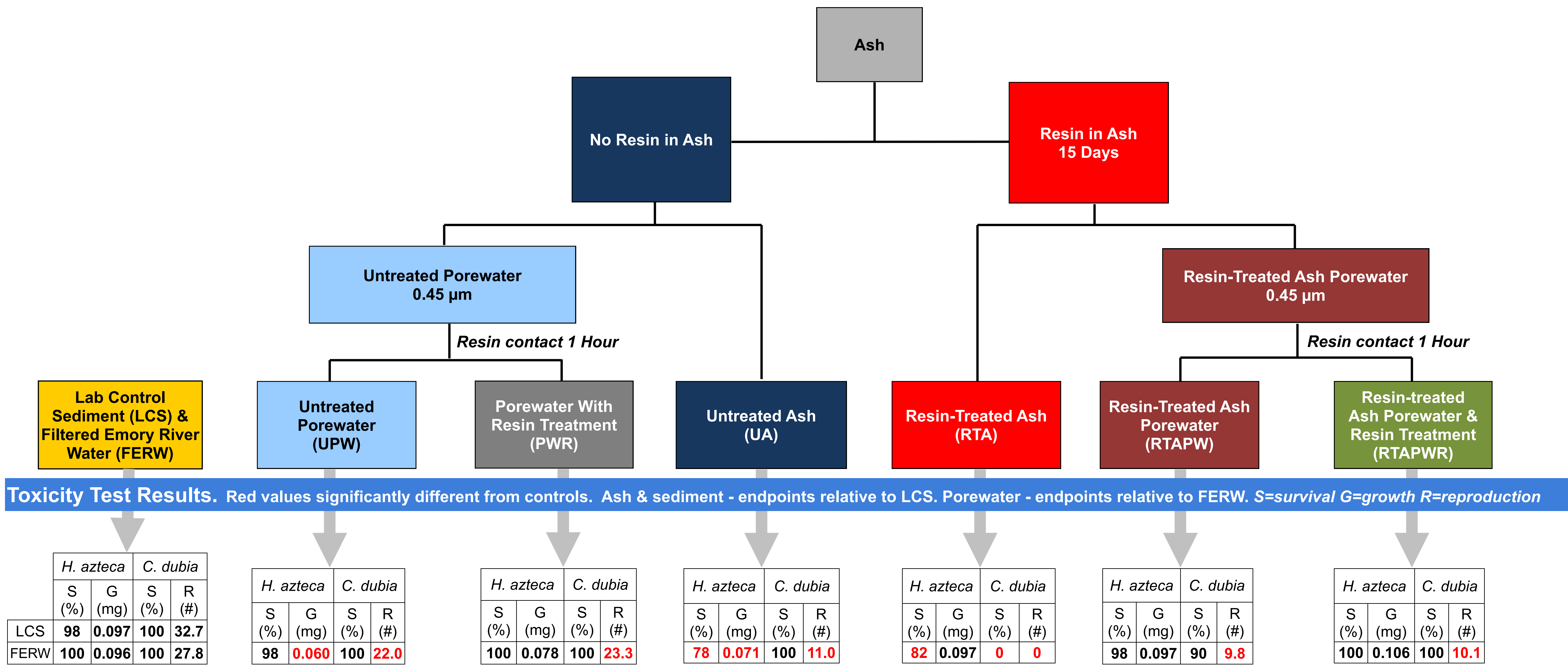
## MATERIALS AND METHODS

- Whole ash sample collected March 18, 2010 from embayment area near failed dredge cell at Kingston Fossil Plant, stored at 0 - 6° C prior to use
- Ash sample: 77.9% solids, 2,650 specific gravity, of which 86% was fly ash, grain size distribution 7.5% sand, 68.4% silt, 24.1% clay, and TOC = 540 mg/kg; acid volatile sulfides not detected
- Laboratory control sediment sample collected March 24, 2010 from Clinch River Mile 189.0, stored at 0 - 6° C prior to use
- Reference control water collected from Emory River Mile 12.2 on April 13, 2010, stored at 0 - 6° C prior to use
- 15 days prior to test start, ash sample homogenized, split into 2 aliquots
- 1 aliquot returned to cold storage for 15 days before further handling/treatment
- 1 aliquot mixed with ResinTech® ASM 10-HP at 20% by weight, returned to cold storage for 15 days
- Day -1 of tests, 2 whole ash aliquots further processed to provide exposure media consisting of UA, UPW, PWR, RTA, RTAPW, RTAPWR
- Porewater obtained via gravity separation during 15-d storage period, filtered with 0.45  $\mu\text{m}$  prior to further handling/treatment. For PWR & RTAPWR, resin added at 10% by weight
- 10-d screening tests with *H. azteca* conducted per EPA Method 100.1
- 7-d screening tests with *C. dubia* conducted per ASTM E 1706-05 Annex A2 and EPA method 1002.0
- ToxCalc™v5.0.23F used for statistical comparisons
- Sediment Total Metals by EPA Method 6010B
- Surface Water Metals by EPA Methods 200.7, 200.8, 7470A/7471A
- Metals analyzed for whole ash (0 - & 10-d), overlying water, porewater
- Decision Matrix developed to evaluate likely causes of measured effects

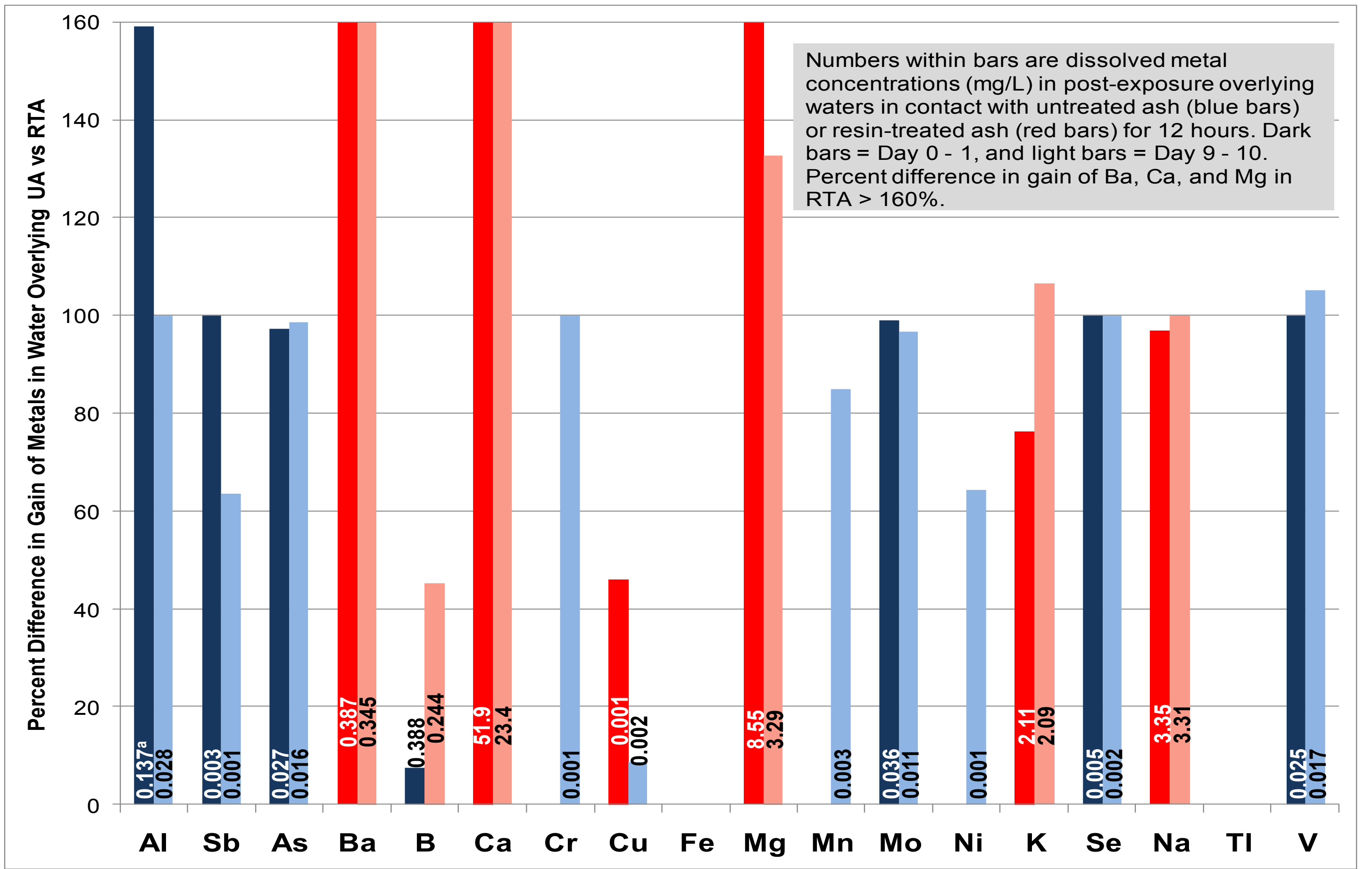
## RESULTS

### Experimental Design of Ash & Porewater Bioavailability Study: *Hyalella azteca* and *Ceriodaphnia dubia*

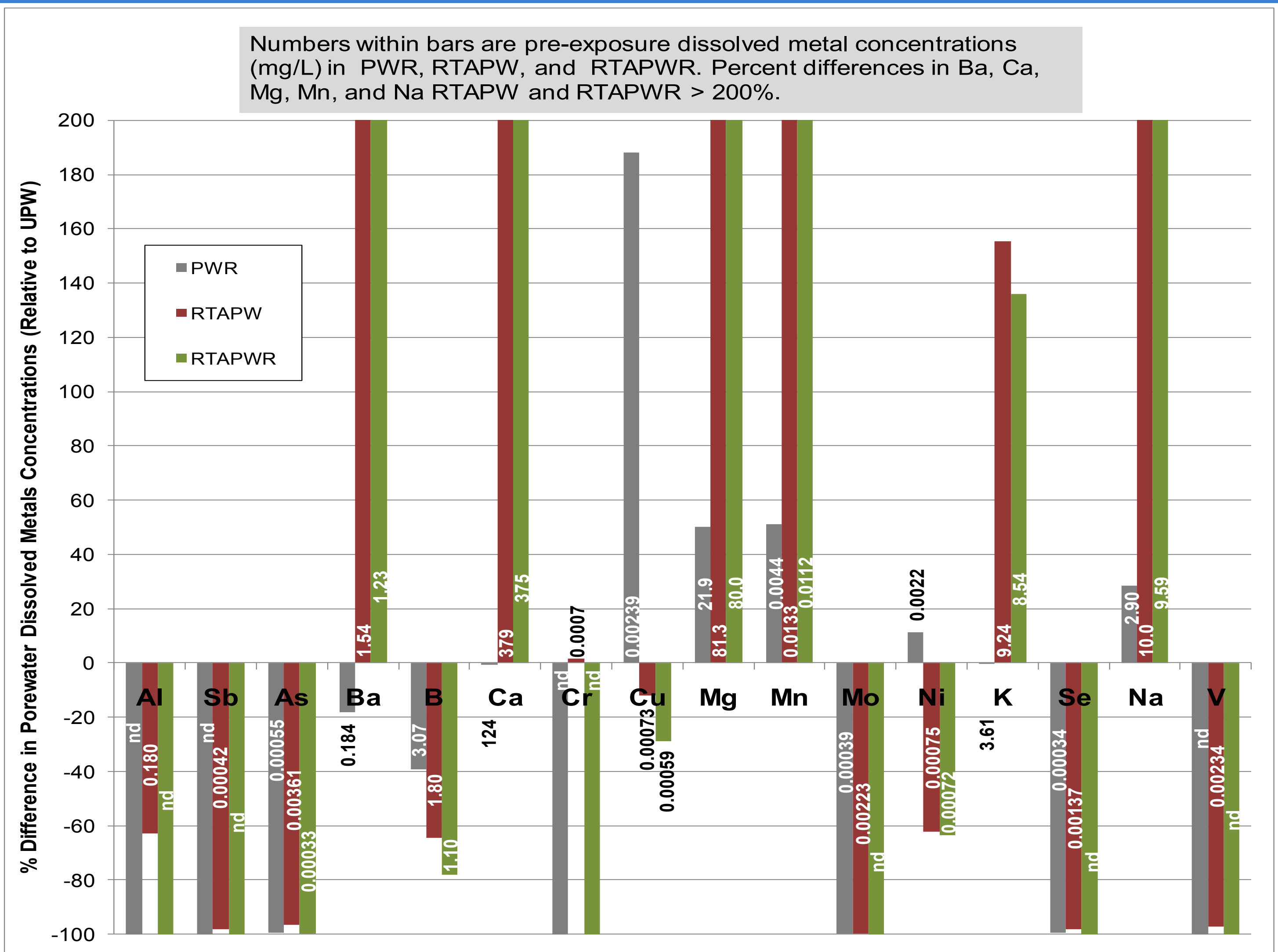
**Note: one-half of the ash sample received no prior treatment before storage at 4°C for 15 days. The other half was mixed with resin (20% by weight) before storage. Porewater (supernatant) was poured off after storage and treated as indicated in the diagram the day before test initiation. Test exposures with the 6 final ash and porewater aliquots began on the same day.**



### Partitioning of Metals to Overlying Water



### Partitioning of Metals to Porewater



## DISCUSSION

### Decision Matrix for Evaluating Likely Causes of Measured Effects

Exposure Media	Endpoint Comparisons	Endpoint Results	Endpoints					
			Survival			Growth		
Ash	UA vs LCS	UA < LCS UA = LCS UA > LCS	x	x	x <sup>1</sup>	x	x	x <sup>1</sup>
	RTA vs LCS	RTA < LCS RTA = LCS RTA > LCS	x	x	x <sup>2</sup>	x	x	x <sup>2</sup>
	UA vs RTA	UA < RTA UA = RTA UA > RTA	x	x	x <sup>2</sup>	x	x	x <sup>2</sup>
Ash/ Porewater	UA vs UPW	UA < UPW UA = UPW UA > UPW	x	x	x <sup>1</sup>	x	x	x <sup>1</sup>
	RTA vs RTAPW	RTA < RTAPW RTA = RTAPW RTA > RTAPW	x	x	x <sup>2</sup>	x	x	x <sup>2</sup>
Porewater	UPW vs FERW	UPW < FERW UPW = FERW UPW > FERW	x	x	x <sup>1</sup>	x	x	x <sup>1</sup>
	UPW vs PWR	UPW < PWR UPW = PWR UPW > PWR	x	x	x <sup>2</sup>	x	x	x <sup>2</sup>
	UPW vs RTAPW	UPW < RTAPW UPW = RTAPW UPW > RTAPW	x	x	x <sup>2</sup>	x	x	x <sup>2</sup>
	UPW vs RTAPWR	UPW < RTAPWR UPW = RTAPWR UPW > RTAPWR	x	x	x <sup>2</sup>	x	x	x <sup>2</sup>

Note: Black letters are expected responses, red letters are study responses based on 2-tailed statistical analyses.

<sup>1</sup> NSD = no significant difference in survival and/or growth between exposure results. No further evaluation needed if endpoints are NSD for both UA vs LCS and UPW vs FERW.

<sup>2</sup> Resin treatment artifact identified. Ash treated with resin produces porewater with high conductivity (up to 3500  $\mu\text{mhos}$ ) and high hardness (up to 1,370 mg/L). *H. azteca* are salt-tolerant organisms, but *C. dubia* exhibit adverse effects when exposed to ionic levels this high.

Treatment artifact observed for *C. dubia* test exposures. Therefore, Decision Matrix for this species is not presented.

## CONCLUSIONS

- Anionic exchange resin treatment of fly ash does not eliminate survival effects of *H. azteca* caused by exposures to whole ash, but does eliminate growth effects
- Metals concentrations in resin-treated whole ash decrease only slightly, but continued loss of Sb, As, and Se is greater in resin-treated ash relative to untreated ash during 10-d exposures to *H. azteca*
- Partitioning of Al, Sb, As, Mo, Se, and V to both overlying water and porewater in untreated ash exposures is greater than in resin-treated ash exposures
- Partitioning of Ba, Ca, Mg, K, and Na to both overlying water and porewater in resin-treated ash exposures is greater than in untreated ash exposures
- Resin treatment increased water conductivities to 3500  $\mu\text{mhos}$  and hardness values to 1,370 mg/L, which were later confirmed to be an artifact of the resin preparation protocol
- In *C. dubia* exposures, these increases in ionic concentration are likely to have caused excessive mortality and decreases in reproduction
- The weight of evidence from this study indicates that *H. azteca* exposures to fly ash result in reduced survival due to physical properties of fly ash, and reduced growth due to metal/metalloid constituents
- No single metal is likely present in concentration sufficient to exert effects on growth, but growth could be affected by a combination of metals that are removed by anionic exchange resins